

Complete Listing of the Claims

✓
Claims 1-63 (Cancelled).

64. (New) A method of making one or more cDNA molecules, comprising:

(a) mixing one or more RNA molecules with (i) one or more polypeptides having reverse transcriptase activity and (ii) at least one primer-adaptor nucleic acid molecule wherein the at least one primer-adaptor nucleic acid molecule comprises one or more ligands and one or more cleavage sites, to form a mixture;

(b) incubating the mixture under conditions sufficient to make one or more cDNA molecules, wherein the one or more cDNA molecules comprise at least one primer-adaptor nucleic acid molecule;

(c) contacting the cDNA molecules with a hapten to produce one or more hapten-cDNA molecule complexes; and

(d) inserting or ligating the cDNA molecules into one or more vectors.

65. (New) The method according to claim 64, further comprising isolating one or more of the hapten-cDNA molecule complexes.

66. (New) The method according to claim 64, wherein the hapten is bound to a solid support.

67. (New) The method according to claim 66, wherein the solid support is selected from the group consisting of nitrocellulose, diazocellulose, glass, polystyrene, polyvinylchloride,

polypropylene, polyethylene, dextran, Sepharose, agar, starch, nylon, beads and microtitre plates.

68. (New) The method according to claim 66, wherein the solid support is a magnetic bead.

69. (New) The method according to claim 64, further comprising contacting the complexes with a restriction enzyme to cleave one or more of the cDNA molecules from the complexes.

70. (New) The method according to claim 69, wherein the restriction enzyme is *NotI*.

71. (New) The method according to claim 69, wherein the cleaved cDNA molecules comprise one sticky end and one blunt end.

72. (New) The method according to claim 71, wherein the sticky end is a *NotI* sticky end and the vector has a *NotI* compatible end and a blunt end.

73. (New) The method according to claim 64, wherein the one or more polypeptides are selected from the group consisting of a Moloney Leukemia Virus (M-MLV) reverse transcriptase, a Rous Sarcoma Virus (RSV) reverse transcriptase, an Avian Myeloblastosis Virus (AMV) reverse transcriptase, a Rous Associated Virus (RAV) reverse transcriptase, a Myeloblastosis Associated Virus (MAV) reverse transcriptase, a Human Immunodeficiency Virus (HIV) reverse transcriptase, a retroviral reverse transcriptase, a

retrotransposon reverse transcriptase, a hepatitis B virus reverse transcriptase, a cauliflower mosaic virus reverse transcriptase, a bacterial reverse transcriptase, and mutants and variants thereof that are substantially reduced in RNase H activity.

74. (New) The method according to claim 64, wherein the conditions sufficient to make one or more cDNA molecules comprise a one or more DNA polymerases, one or more nucleotides and one or more primers.

75. (New) The method according to claim 74, wherein the primers are primer-adapters that comprise one or more ligands and one or more cleavage sites.

76. (New) The method according to claim 64, wherein at least one of the RNA molecules is an mRNA molecule.

77. (New) The method according to claim 64, wherein at least one of the RNA molecules is polyadenylated.

78. (New) The method according to claim 64, wherein the one or more RNA molecules is a population of RNA molecules.

79. (New) A method of making a cDNA molecule, comprising

(a) mixing one or more RNA molecules with (i) one or more polypeptides having reverse transcriptase activity and (ii) at least one primer-adaptor nucleic

acid molecule wherein the at least one primer-adapter nucleic acid molecule comprises a restriction enzyme recognition sequence and at least one biotin moiety, to form a mixture;

(b) incubating the mixture under conditions sufficient to make one or more cDNA molecules, wherein the one or more cDNA molecules comprise at least one primer-adapter nucleic acid molecule;

(c) contacting the cDNA molecules with one or more solid supports to which are bound avidin and/or streptavidin, to produce one or more solid support-cDNA molecule complexes;

(d) contacting the complexes with a restriction enzyme that cleaves the restriction enzyme recognition sequence in the adapter-primer; and

(d) inserting or ligating the cDNA molecules into vectors.

80. (New) The method according to claim 79, further comprising isolating one or more of the solid support-cDNA molecule complexes.

81. (New) The method according to claim 79, wherein the solid support is selected from the group consisting of nitrocellulose, diazocellulose, glass, polystyrene, polyvinylchloride, polypropylene, polyethylene, dextran, Sepharose, agar, starch, nylon, beads and microtitre plates.

82. (New) The method according to claim 79, wherein the solid support is a magnetic bead.

83. (New) The method according to claim 79, wherein the restriction enzyme is *NotI*.

84. (New) The method according to claim 79, wherein the one or more polypeptides are selected from the group consisting of a Moloney Leukemia Virus (M-MLV) reverse transcriptase, a Rous Sarcoma Virus (RSV) reverse transcriptase, an Avian Myeloblastosis Virus (AMV) reverse transcriptase, a Rous Associated Virus (RAV) reverse transcriptase, a Myeloblastosis Associated Virus (MAV) reverse transcriptase, a Human Immunodeficiency Virus (HIV) reverse transcriptase, a retroviral reverse transcriptase, a retrotransposon reverse transcriptase, a hepatitis B virus reverse transcriptase, a cauliflower mosaic virus reverse transcriptase, a bacterial reverse transcriptase, and mutants and variants thereof that are substantially reduced in RNase H activity.

FT
COPY
SUB
G1
85. (New) The method according to claim 79, wherein the conditions sufficient to make one or more cDNA molecules comprise one or more DNA polymerases, one or more nucleotides and one or more primers.

86. (New) The method according to claim 79, wherein at least one of the RNA molecules is an mRNA molecule.

87. (New) The method according to claim 79, wherein at least one of the RNA molecules is polyadenylated.

88. The method according to claim 79, wherein the one or more RNA molecules is a population of RNA molecules.

89. (New) A method of making one or more cDNA molecules, comprising:
- (a) mixing one or more mRNA molecules with (i) one or more polypeptides having reverse transcriptase activity and (ii) at least one primer-adaptor nucleic acid molecule, wherein the at least one primer-adaptor nucleic acid molecule comprises one or more ligands and one or more cleavage sites, to form a mixture; and
 - (b) incubating the mixture under conditions sufficient to make one or more double stranded cDNA molecules, wherein the one or more cDNA molecules comprise at least one primer-adaptor nucleic acid molecule;
 - (c) contacting the mixture with a hapten under conditions sufficient to form one or more hapten-cDNA molecule complexes; and
 - (d) isolating one or more of the complexes comprising the cDNA molecules.
90. (New) The method according to claim 89, further comprising digesting the complexes with a single restriction enzyme that cleaves one or more of the complexes at cleavage sites in the primer-adaptors to produce cleaved cDNA molecules.
91. (New) The method according to claim 90, further comprising inserting or ligating the cleaved cDNA molecules into vectors.
92. (New) The method according to claim 89, wherein the hapten is bound to a solid support.
93. (New) The method according to claim 92, wherein the solid support is selected from the group consisting of nitrocellulose, diazocellulose, glass, polystyrene, polyvinylchloride,

polypropylene, polyethylene, dextran, Sepharose, agar, starch, nylon, beads and microtitre plates.

94. (New) The method according to claim 92, wherein the solid support is a magnetic bead.

95. (New) The method according to claim 91, wherein the restriction enzyme is *NotI*.

96. (New) The method according to claim 91, wherein the cleaved cDNA molecules comprise one sticky end and one blunt end.

97. (New) The method according to claim 96, wherein the sticky end is a *NotI* sticky end and the vector has a *NotI* compatible end and a blunt end.

98. (New) The method according to claim 89, wherein the one or more polypeptides are selected from the group consisting of a Moloney Leukemia Virus (M-MLV) reverse transcriptase, a Rous Sarcoma Virus (RSV) reverse transcriptase, an Avian Myeloblastosis Virus (AMV) reverse transcriptase, a Rous Associated Virus (RAV) reverse transcriptase, a Myeloblastosis Associated Virus (MAV) reverse transcriptase, a Human Immunodeficiency Virus (HIV) reverse transcriptase, a retroviral reverse transcriptase, a retrotransposon reverse transcriptase, a hepatitis B virus reverse transcriptase, a cauliflower mosaic virus reverse transcriptase, a bacterial reverse transcriptase, and mutants and variants thereof that are substantially reduced in RNase H activity.

99. (New) The method according to claim 89, wherein the conditions sufficient to make one or more cDNA molecules comprise one or more DNA polymerases, one or more nucleotides and one or more primers.

100. (New) The method according to claim 99, wherein the primers are primer-adapters that comprise one or more ligands and one or more cleavage sites.

101. (New) The method according to claim 89, wherein at least one of the RNA molecules is an mRNA molecule.

102. (New) The method according to claim 89, wherein at least one of the RNA molecules is polyadenylated.

103. (New) The method according to claim 89, wherein the one or more RNA molecules is a population of RNA molecules.

104. (New) A method of making one or more cDNA molecules, comprising:

(a) mixing one or more mRNA molecules with (i) one or more polypeptides having reverse transcriptase activity and (ii) at least one primer-adaptor nucleic acid molecule, wherein the at least one primer-adaptor nucleic acid molecule comprises one or more ligands and one or more cleavage sites, to form a mixture; and

(b) incubating the mixture under conditions sufficient to make one or more double stranded cDNA molecules, wherein the one or more cDNA molecules comprise at least one primer-adaptor nucleic acid molecule;

(c) contacting the mixture with a hapten under conditions sufficient to form one or more hapten-cDNA molecule complexes; and

(d) digesting the complexes with a single restriction enzyme that cleaves one or more of the complexes at cleavage sites in the primer-adapters to produce one or more cleaved cDNA molecules.

105. (New) The method according to claim 104, further comprising isolating one or more of the complexes.

106. (New) The method according to claim 104, further comprising ligating or inserting the cleaved cDNA molecules into vectors.

107. (New) The method according to claim 104, wherein the hapten is bound to a solid support.

108. (New) The method according to claim 108, wherein the solid support is selected from the group consisting of nitrocellulose, diazocellulose, glass, polystyrene, polyvinylchloride, polypropylene, polyethylene, dextran, Sepharose, agar, starch, nylon, beads and microtitre plates.

109. (New) The method according to claim 108, wherein the solid support is a magnetic bead.

110. (New) The method according to claim 104, wherein the restriction enzyme is *NorI*.

111. (New) The method according to claim 106, wherein the cleaved cDNA molecules comprise one sticky end and one blunt end.
112. (New) The method according to claim 111, wherein the sticky end is a *NotI* sticky end and the vector has a *NotI* compatible end and a blunt end.
113. (New) The method according to claim 104, wherein the one or more polypeptides are selected from the group consisting of a Moloney Leukemia Virus (M-MLV) reverse transcriptase, a Rous Sarcoma Virus (RSV) reverse transcriptase, an Avian Myeloblastosis Virus (AMV) reverse transcriptase, a Rous Associated Virus (RAV) reverse transcriptase, a Myeloblastosis Associated Virus (MAV) reverse transcriptase, a Human Immunodeficiency Virus (HIV) reverse transcriptase, a retroviral reverse transcriptase, a retrotransposon reverse transcriptase, a hepatitis B virus reverse transcriptase, a cauliflower mosaic virus reverse transcriptase, a bacterial reverse transcriptase, and mutants and variants thereof that are substantially reduced in RNase H activity.
114. (New) The method according to claim 104, wherein the conditions sufficient to make one or more cDNA molecules comprise one or more DNA polymerases, one or more nucleotides and one or more primers.
115. (New) The method according to claim 114, wherein the primers are primer-adapters that comprise one or more ligands and one or more cleavage sites.

116. (New) The method according to claim 104, wherein at least one of the RNA molecules is an mRNA molecule.

117. (New) The method according to claim 104, wherein at least one of the RNA molecules is polyadenylated.

118. (New) The method according to claim 104, wherein the one or more RNA molecules is a population of RNA molecules.

sub
G1
~~FT~~
~~CDOT~~